GREEN SYNTHESIS OF ZINC OXIDE NANOPARTICLE BY MICROBES AND THEIR ROLE IN HEAVY METALS REMEDIATION

A DISSERTATION SUBMITTED FOR THE PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF

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CONDIDATE'S DECLARATION

I, **Manesh**, [**2k21/MSCBIO/23]**, final semester student of M.Sc. Biotechnology, hereby declare that the project Dissertation titled "**Green synthesis of Zinc oxide nanoparticle by microbes and their role in heavy metals remediation**" which is submitted by me to the Development of Biotechnology, Delhi Technological University, Delhi in partial fulfillment of the requirement for the award of the degree of Master of Technology, is original and not copied from any source without proper citation. This work has not previously formed the basis for the award of any Degree, Diploma Associateship, Fellowship or other similar title or recognition.

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CERTIFICATE

I, hereby certify that the Project Dissertation titled, "**Green synthesis of Zinc oxide nanoparticle by microbes and their role in heavy metals remediation"** which is submitted by, **Manesh [2K21/MSCBIO/23]**, Department of Biotechnology, Delhi Technological University, Delhi in partial fulfillment of the requirement for the award of the degree of Master of Science, is a record of the project work carried out by the student under my supervision. To the best of my knowledge this work has not been submitted in part or full for any Degree or Diploma to this University or elsewhere.

Place: Delhi **Prof. Jai Gopal Sharma**

Date: **SUPERVISOR**

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MANESH

ABSTRACT

Microbes and plant extracts can be used to create nanoparticles. Size and surface of nanoparticles can be easily changed under the right parameters to provide both passive and active medication delivery. By mounting ligands to the surface of the nanoparticle, it can be directed towards the targeted spots. The study of particles with at least one nanometer in dimension is known as nanotechnology. There are three ways to create nanoparticles: physically, chemically, and through green synthesis techniques. Plant extracts, enzymes, or microbes can all be used in green synthesis. Due to its non-toxic and environmentally beneficial procedure, the green synthesis method provides the basis for this project activity. In this experiment, E.coli and other *Bacillus* species are used to reduce Zinc (II) ion solution into ZnO nanoparticles. Zinc nanoparticles were characterised using a UV-visible spectrophotometer to determine their Surface Plasmon resonance, a SEM to determine their morphologies, and an XRD and FTIR to determine their crystallinity.

CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF KEYWORDS AND ABBREVIATIONS

SEM.	Scanning electron microscope
UV- visible.	UV visible spectrophotometer
ZnO.	Zinc oxide
NPs.	Nanoparticles
FTIR.	Fourier Transform Infrared Spectroscopy
NMR.	Nuclear Magnetic resonance
GaN.	Gallium nitride
DMF.	N, N-dimethylformamide
MBC.	Minimum bactericidal concentration
ROS	Reactive oxygen species
Ni.	Nickel
MIC.	Minimum inhibitory concentration
SPR.	Surface Plasmon resonance

CHAPTER 1

INTRODUCTION

The benefits of new technology are typically accompanied by new scientific difficulties, health concerns for people, and a lots of environmental problems. The nanoparticle's broad usage in the fields of science and industry holds considerable promise as a result of recent developments in nanotechnology. The most significant development in the area science and technology over the past few decades is due to nanotechnology. Nanotechnology, which refers to the nanoscale, or 1nm to 100 nm, is a modern, well-established technology. Typically inorganic, metal oxide nanoparticles. Numerous nanoparticles, such as Ni, Fe, Mn, Co and Zn, are recognised as the commonly used magnetic materials in electronic ignition systems, vending machines, wrist watches, medical implants, transformer circuits, inductor core, recording equipment, and magnetic sensors, telecommunications, magnetic fluids, and microwave, among other applications. They are also applicable to other high-frequency applications [1, 2]. Nanoparticles have many uses in the medical sciences, including drug delivery, imaging, and diagnostics. Nanoparticles have unique properties such as small size and high surface area to volume ratio. Zinc oxide nanoparticles' ability to fight germs and fungus has been proven in numerous investigations. Various metal nanoparticles, including silver nanoparticles, exhibit these properties as well.

Due to its eco-friendliness to the Nano-industries, more often than other methods, microorganisms are used to produce various nanoparticles. Different kinds of bacterial species have a propensity to synthesize intracellularly both metal nanoparticles and their alloys [3]. Biological processes remove the complex process of Bacterial culture maintenance by using different plant species to make nanoparticles , which are beneficial for our ecosystem.

Because of their distinctive physical characteristics, such as electrical conductivity, optical band gap, refractive index, magnetic properties, and superior mechanical properties, such as hardness of nanomaterials, and chemical properties compared to their counterpart bigger structured materials, Nano sized materials have already been thoroughly studied by researchers worldwide [5].Novel antibacterial agents are another name for nanoscale materials. Due to increasing and developing microbial resistances against various metal ions, various antibiotics, and the development of resistant strains in numerous ways [6], researchers are currently focusing on antimicrobial agents that have a high surface area to volume ratio.

Nanoparticles' size, shape, morphology, composition, and crystallinity all affect their antibacterial activity. Researchers are working to create nanoparticles, Nano devices, which are implemented in a variety of organizations and fields, including food technology, environment, energy, and medical science.

Zinc oxide nanoparticles have a wide range of industrial applications, including photo catalysts, gas sensors, solar cells, UV light emitting devices, pharmaceuticals, and cosmetics [7–11]. t is also referred to as a versatile semiconductor material that has drawn interest and attention for its wide range of applications in many fields, including surface acoustic wave (SAW) devices, UV photodiodes, electrochemical cells, and biological labels. The ZnO nanoparticle is reasonably priced and practical. It has a wurtzite like structure and, it is an n-type semiconductor. Additionally, it exhibits optical transparency in the visible spectrum. For white-light LEDs, zinc oxide's ability to emit visible light is extremely important [12].This nanoparticle is made of metal oxide. The metal nanoparticles are non-toxic, self-cleaning, and have surface Plasmon resonance characteristics in the UV-visible region [13]. ZnO has a potent antibacterial effect and is harmless. Both gram-positive and gram-negative bacteria are susceptible to the bactericidal actions of ZnO nanoparticles [14]. Additionally, they show antibacterial efficacy against high-temperature and high-pressure-resistant spores. The antibacterial agents are substances, either natural or artificial, which are useful in preventing microbial growth. The majority of the antimicrobial substances utilised in the textile sector are biocides.

CHAPTER 2

LITERATURE OF REVIEW

2.1 Nanotechnology and Nanoscience

Since nanotechnology has so many applications in research technology and other branches of medical science, it is regarded as a Topic that is still developing. . The term “Nano” derives from a Greek term that means “extremely small [15].The physicist Richard Feynman presented the fundamental ideas of nanoscience and nanotechnology on December 29, 1959, at the California Institute of Technology (CalTech), under the title “There’s a Plenty of Room at the Bottom” [16]. Professor Norio Taniguchi eventually came up with the name “Nanotechnology” by drawing on Feynman’s investigations into ultra-precise machining. The phrase "nanotechnology" was coined by Nori Taniguchi in 1974 while he was speaking at the Tokyo International Conference about manufacturing engineering. His studies focused on the working of ultrasonic machining of brittle, hard materials like quartz crystals, alumina, and silicon ceramics. He focused in his talk on ultra-precise machining. Richard Feynman stated in a lecture that the SEM will gain a precision and stability, thereby rendering it is valuable and feasible for all to "see" atoms. In order to properly detect and identify individual atoms, scanning tunneling microscopes, which were first created in 1981, are helpful. Feynman went on to say that it is possible to organize atoms in small structures that are chemically stable, which would create materials at the molecular and atomic levels.

2.2 Nanomaterials

The state of matter at the nanoscale is known as nanomaterials. They are considered as a functional unit of nanotechnology. Organics, metals, metal oxides, semiconductors, and carbon-based materials are the “Building Blocks” for nanomaterials. Thin films, cluster, multilayers, and Nanocrystalline materials all are the instances of nanomaterials with structural characteristics at the nanometer. Nanomaterials are classified based on the dimensions of 0, 1, 2, and 3. Materials in the form of thin films, cluster, bulk Nanocrystalline and multilayer materials, among others, are amorphous, metals and, semiconductors, crystalline alloys, carbide ceramics and oxides etc.

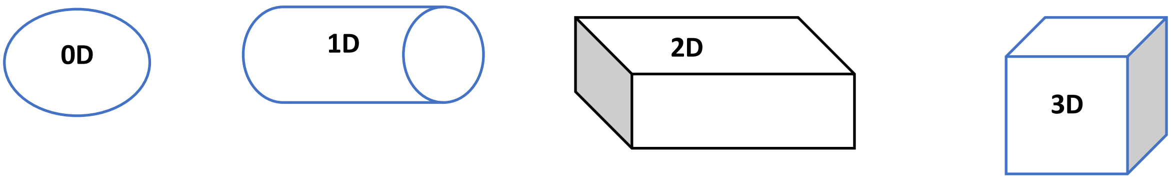


Fig.1 Classification of Nanoparticles

Table.1. Categories based on dimension

Dimension	**Nanomaterials examples**
0D	Nps, nanocluster, nanodots
1D	Nanotubes, nanowires, Nano rods
2D	Membrane, quantum wells
3D	Block polymer, nanocrystal arrays

Carbon black, a well-known nanomaterial, was first used in industrial production more than a century before. Fumed silica, a kind of titanium dioxide (TiO2), dioxide (SiO2), and zinc oxide (ZnO) are the other early nanomaterials. The existence of different properties for nanomaterials at the nanoscale is further demonstrated by the quantum effects. All nanomaterials have different magnetic properties, refraction of light, increased heat and electricity, electrical conductivity, and even colour changes in response to size variation.

Nanocrystalline nanomaterials, which are less than 100 nm in size, are controlled to display atom-like behaviour. They also have a high surface energy because they have a larger surface area and a wider band gap between the valence and conduction bands. It happens when they are split almost into atomic pieces [17, 18]. ‘’A wonder of modern medicine’’ is another term used to describe nanomaterials. If nanomaterials can kill at least 650 cells, it emphasises the value of antibiotics that eradicate at least six different disease-causing organisms [19].Research on nanomaterials is thriving for specialised applications such microbial growth suppression, antibiotic carriers, and death agents [20].

2.3. Metal oxide nanoparticles

The production of various nanoparticle by physical and chemical processes is not appropriate. These methods typically include the use of hazardous chemicals, and the elevated temperatures that aid in the production of the corresponding nanoparticles which are harmful to the environment [21, 22]. Additionally, metal oxides have been used as sorbents for a number of environmental contaminants. Physical and chemical synthesis are inferior to biological synthesis since it is more environmentally friendly. The biological methods for synthesizing nanoparticles have been proposed as the likely and promising environmental friendly alternatives to the chemical and physical methods of synthesis [23]. These methods are based on microbes, enzymes, plants, and their extracts. For particular biomedical applications and studies, nanoparticle biocompatibility is especially crucial [24].

2.4 Synthesis of nanoparticles

For the quick and easy synthesis of huge numbers of ZnO nanoparticles, there are a number of physical and chemical processes. Sol-gel, solvothermal, Chemical precipitation, hydrothermal, photochemical and electrochemical reduction procedures are among the simple-solution based techniques that are preferred. Using green synthesis techniques, ZnO-NPs can be produced from various plant species' leaf extracts as well as cellular extracts from bacteria and fungus. Various enzymes also control the environmental friendly creation of nanoparticles. ZnO nanostructures can be created using a types of techniques, including high temperature evaporation, organic vapour phase epitaxy, pulsed laser deposition, gas spraying, sol-gel, sputtering, electrochemical and wet chemical processes. Applications for nanoparticles vary depending on the numerous production procedures that have been developed [25]. The following are the several techniques used to create nanoparticles:

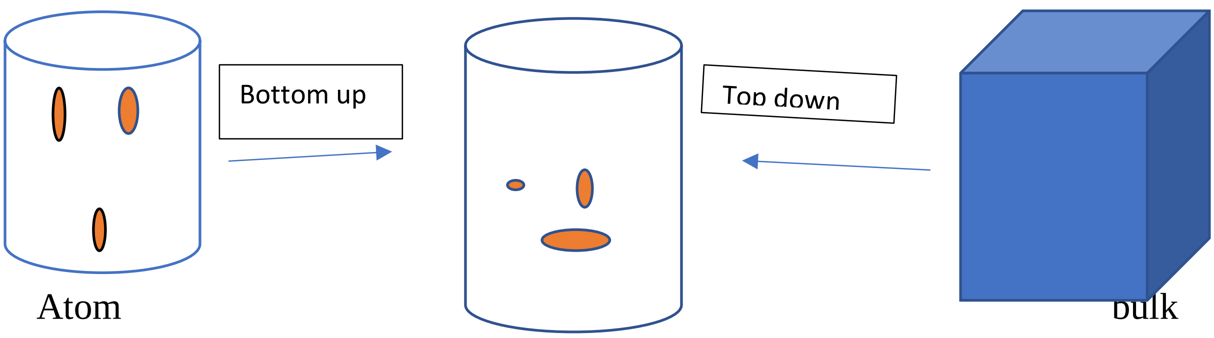
- Physical synthesis process
- Chemical synthesis process
- Biological synthesis process

2.4.1 Physical method

Physical processes used to create ZnO nanoparticles frequently call for specialised tools or high operational control. The physical approach, which is frequently referred to as top-down, involves techniques including diffusion, irradiation, thermal breakdown, and arc discharge, among others. One important top-down strategy is the thermal decomposition method. It is employed to make nondispersive nanoparticles. These methods begin the processing of nanostructures using larger starting structures or macroscopic units that can be controlled from the outside. Physical top-down methods of synthesizing nanoparticles include etching through a mask, ball milling, and applying extreme plastic deformation. A model that generates a larger atom for subsequent nanoscale reduction is the top-down approach of synthesis. Most often, it takes longer and costs more money to produce top-down methods. Due to its slowness, this technology cannot be used for mass production.

2.4.2. Chemical method

The majority of chemical methods for creating nanoparticles are bottom-up strategies. These techniques require the atomic-scale reduction of substances parts, followed by steps that produce nanostructures by self-assembly. Basic building blocks unite through the self-assembly of nanoscale physical forces into more substantial and stable structures. The production of quantum dots and nanoparticles from colloidal dispersion are two examples of this. These processes result in the reduction of various metal particles to nanoparticles. Different kinds of metal particles, including sodium citrate and sodium borohydrate, are employed [56]. N, N-dimethylformamide (DMF), Poly (N-vinyl pyrrolidine (PVP), ethyl alcohol, tetra-n-tetra-fluoroborate (TFATEB), CTAB, and other chemical agents are also employed [18, 19, 20].



Chemical reduction

Fig.2. methods of synthesis

2.4.3. Biological method

The green synthesis approach has been shown to be superior to previous methods used for the creation of nanoparticles. Due to the absence of harmful ingredients, green synthesis techniques are environmentally safe and suitable for pharmaceutical and other biomedical applications [26]. Additionally, this approach doesn't need high pressure or temperature. Due to its eco-friendliness, microbes are utilised more frequently than other processes to create different nanoparticles. The manufacture of zinc nanoparticles using ecologically friendly components including plant leaf extract, bacteria, fungus, and enzymes offers a wealth of advantages in terms of Eco friendliness and compatibility for pharmaceutical and many other biological applications [2].

2.5. ZnO nanoparticles

The inorganic compound zinc oxide has the formula of ZnO. It's a white colored powder that won't dissolve in water. By using new atomistic potentials, the configuration and the structural components of ZnO-NPs have been studied. For various uses of nanoparticles, mechanical qualities such internal stress and adhesion characteristics must be maintained in order to preserve durability and patterning accuracy. The structure of the ZnO is wurtzite hexagons. ZnO often crystallizes as a mix of zinc and hexagonal wurtzite. With a significant exciting binding energy of 60 meV and a broad band gap of 3.37 eV, zinc oxide has recently been recognised as a crucial semiconductor [27].

2.6. Properties of nanoparticle

Nanometer-sized semiconductors are significant because of their optical, electrical and chemical characteristics, which may be customized by varying particle size.

The following categories apply to the characteristics of zinc oxide nanoparticles:

- Physical property
- Electrical property
- Mechanical property
- Chemical property

6.2.1 Physical property

Zinc oxide is colorless and has a whitish appearance in its pure microcrystalline state. When heated, zinc oxide turns yellow, and when cooled, it returns to its natural colour of white.The melting point of zinc oxide is over 1200 degrees Celsius at atmospheric pressure, while it is over 1975 degrees Celsius at high pressure. At a temperature of 1500 C, the vapour pressure is 12 mm., the heat of sublimation is 129 Kcal/mole when the vapour is not disassociated and 193

Kcal/mole when the vapour is associated. The heat Capacity (Cp) is 9.62 cal/deg/mole and thermal expansion coefficient is 4 x 10(-6)/°Cat 25 °C.

2.6.2. Electrical property

For the advancement of ZnO nanostructures' potential uses in nanoelectronics, it is essential to conduct fundamental research on their electrical characteristics. At ambient temperature, ZnO has a band gap that is 3.3 eV, which is moderately large value. The benefits of having a large band gap include the capacity to operate at high temperatures and powers, as well as higher breakdown voltages. When doping is not present, ZnO has an n-type character. The n-type property

Is frequently caused by the non-stoichiometry analysis. There is a Oxygen vacancies, zinc interstitials site and ZnO nanowires cause to display n-type Semiconductor behaviour.

2.6.3. Mechanical Property

In comparison to GaN, which has an exciting energy of about 23 meV, ZnO exhibits a number of advantages, including having an exciting energy of about 60 meV. Effective laser UV applications can exploit this. Zn oxide has the greatest piezoelectric tensor among all semiconductor. This will provide it an important and necessary property for many piezoelectric applications that need for a strong electro-mechanical interaction between them.

2.6.4. Chemical Property

The mineral zincates and the white powder known as zinc white are both forms of zinc oxide. It usually has an orange or red tint because of manganese impurities. Natural thermochromism occurs in crystalline zinc oxide, which turns from white to yellow when heated and back to white when cooled. At high temperatures, a small oxygen loss causes this change in colour.

2.7. Applications of ZnO nanoparticles

- It has been utilised as a source of zinc, a trace metal that is crucial to the food sector [28].
- It is now widely acknowledged that as compared to smaller particles, nanoparticles more effectively permeate cell membranes. Few studies, however, have examined how ZnO nanoparticles' physicochemical properties affect cellular absorption, which could reveal important details about their potential toxicity.
- ROS are present on the surface of ZnO-NPs, which gives them their remarkable resistance to microbes.
- The main benefits of ZnO are its inexpensive cost, excellent gas sensing capabilities, photo catalytic activity, antibacterial activity, and the ability

to create structures with unique optical features, such as photonic crystals and catalytic materials. [29].

2.8. Minimum inhibitory concentration

The term "minimum bactericidal concentration" (MBC) define to the lowest antimicrobial agent concentration that, following subculture on to antibiotic-free media, stops the development of a certain pathogen. The term "minimum inhibitory concentration" refers to the lowest antimicrobial agent level that, after an overnight incubation, reduces a microorganism's ability to grow visibly. The main purpose of MIC in diagnostic labs is to validate resistance. However, the majority are research instruments for figuring out new antimicrobials’ in vitro activity and using the information from that study to figure out MIC break points [30].The minimum inhibitory concentration test determines a material’s antibacterial property both for general microorganisms and against specific bacteria-like pathogens.

2.9. Antimicrobial activity

ZnO, a metal oxide powder, exhibits very strong growth inhibitory inhibition of a wide spectrum of microorganisms. Although other processes have also been proposed, the main catalyst for the synthesis of the reactive oxygen species (ROS) from oxygen and water that compromise the integrity of the bacterial cell membrane is the mechanism for ZnO’s antibacterial activity. Particles with more surface area exhibit better antibacterial activity because the process of radical generation takes place on the particle surface. The size of the ZnO particles consequently decreases with increased antibacterial activity.

2.10. Possible types of characterization methods

- Electron probe techniques
- Spectroscopic techniques
- Scanning probe techniques
- Ion–particle techniques

CHAPTER 3

MATERIAL AND METHODS

3. Bacterial strain

S aureus, E. coli, Klebsiella pneumonia, and Bacillus species are some of the bacteria that can cause respiratory infections.

3.2 Glassware and apparatus

Everything made of glass, including, beakers, measuring cylinders conical flasks, funnels, 96-well plates, test tubes, Petri plates, filter paper (Whatman 40), microfuge tips and eppendorf tubes was bought from Borosil in India.

3.3 Chemicals

Zinc acetate dehydrate (4.4g), 1M NaOH (10ml), ethanol (25ml)

3.4 Synthesis of Nanoparticles

3.4.1 Extract preparation

- 100 ml of 24 days old culture was centrifuged at 10000 rpm for 5 minutes.
- The pellet was rinsed with DW two to three times after the supernatant was removed.
- After being thoroughly cleaned, the pellet was dissolved in distilled water of 100 ml and left at 80°C for 30 minutes.

- Cooling down of the mixture, supernatant (bacterial extract) was collected using Centrifugation.
- The prepared extract was stored at 4°C for further use.

3.4.2 Method of preparation

Zn-NPs were prepared by the addition of a proportion of 10 mM solution of AgNO3 in Algal extract and incubating the obtained mixture at a particular pH and temperature.

3.4.3 ZnO NP synthesis using cell free extract

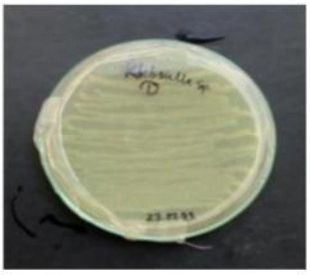
- 100 ml of 24 days old culture was centrifuged at 10000 rpm for 5 minutes and the Supernatant was collected in a separate flask
- The cell free extract was maintained at pH 5-10.
- 10mM fresh Zinc acetate solution was prepared and mixed with the cell free extract Maintained at pH 5-10 in the ratio of 1:10 (Precursor metal salt: Bacterial extract).
- 0 min absorbance was scanned (200 nm- 700nm) using UV-Visible spectrophotometer.
- The mixture was incubated at 28oC, still in light conditions for 24 hours.
- Color change was observed and absorbance scan was run for all the samples at 0 hrs., 24 hrs., 48 hrs. and 96 hrs. for checking the stability of produced ZnO-NPs.

- BG11 medium and 1g/L glucose solutions were also mixed with 10 mM Zinc acetate in the Ratio of 1:10 (Precursor metal salt: solution) to check their ability to synthesize ZnO-NPs.

3.5 Culture preparation for antimicrobial test

Against *E. coli*, *Salmonella Sp*, *Bacillus cereus*, *Staphylococcus aureus*, and *Klebsiella sp*., the produced ZnO-NPs were evaluated in triplicates for their antibacterial activity.

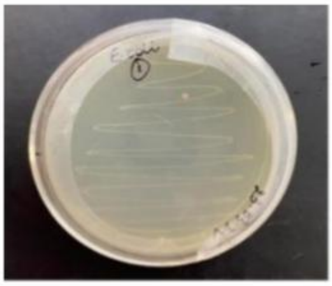
- On the nutrient Agar plate, 0.011 ml of overnight-grown bacterial culture was applied using a spreader.
- Filter paper was cut into discs using an aseptic process.
- Different amounts of the synthesized ZnO-NPs solution (10, 20, 30, 40, and 50 L) were absorbed into the filter-paper discs before they were carefully placed on the nutrient agar plates with the cultures.
- Overnight incubation carried out for 24 hours.
- A millimeter scale was used to carefully measure the plates and look for any zones of restriction around the discs.



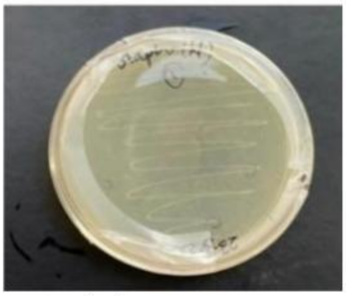
Klebsiella sp

Bacillus cereus.

Salmonella sp



E.coli.



staphylococcus aureus

Fig. 3 Nutrient Agar plate containing different Bacterial species

3.6 Characterization of ZnO nanoparticles

The ZnO NPs produced by *E. coli* were examined using a UV-Visible Spectrophotometer to confirm ZnO-NP formation, a FE-SEM to determine shape, an XRD analysis to determine crystallinity, a Zeta-potential to determine size, and an FTIR to identify bioactive compounds acting as stabilizing and capping agents for ZnO-NPs.

3.7. Heavy metals remediation

3.7.1 Nickel remediation

Testing was done to determine how well the biosynthesized ZnO-NPs removed nickel from synthetic wastewater.

- Nickel standard curves with various concentrations were created, and each dilution's absorbance was recorded.
- Biogenic ZnO NPs were combined at concentrations of 2 mg and 4 mg with a 100 PPM nickel solution that had a pH range of 3 to 7.
- After combining ZnO-NPs with the chromium solution, absorbance was observed.
- The solutions were incubated in a 200 rpm rotating shaker at room temperature for 24 hours.
- After 24 hours of incubation, the solution was centrifuged at 10,000 rpm for 5 minutes, and the absorbance was measured.
- The standard curve was used to determine the removal efficiency.

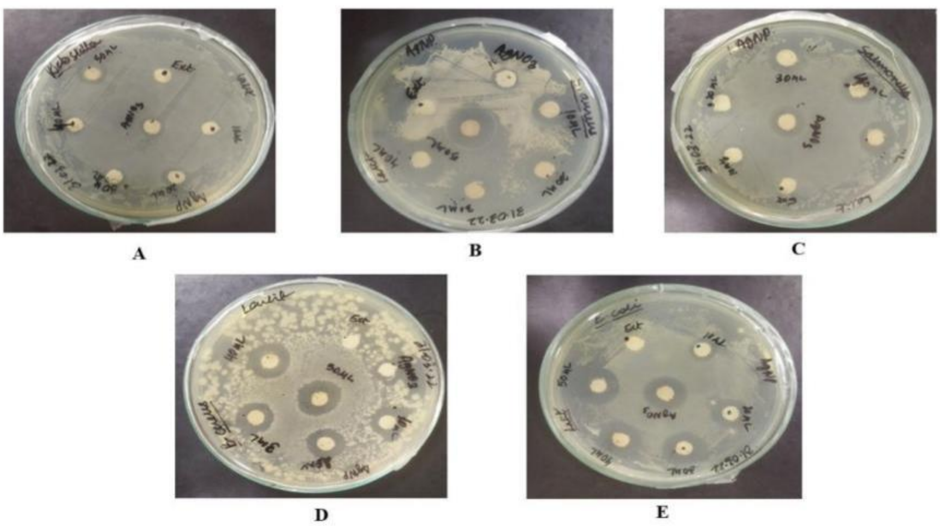
Removal efficiency (%) = [(initial conc. – final conc.)/ Initial conc.] * 100

CHAPTER 4

RESULT AND DISCUSSION

4.1 Antibacterial activity ZnO NP

The zone of inhibition created by various ZnO-NP volumes was used to assess the synthesized ZnO-NPs' potency as an antibacterial agent. Bacterial extract and zinc acetate solution were used as controls, and the zone of inhibition for the test solutions was also examined. Higher concentrations of the synthesized ZnO-NPs were found to be far more potent antibacterial agents than lesser quantities. ZnO-NPs provided the largest zone of inhibition against B. cereus (21 mm) among the five test bacterial species, followed by *E.coli* (20 mm) and *S. aureus* (21 mm).As shown in figure 4, *Klebsiella sp*. Was discovered to be least susceptible to ZnO-NPs while *Salmonella sp*. was found to be only moderately efficient against ZnO-NPs. The existence of an acidic capsule around the bacterial cell wall may have hindered the diffusion of ZnO-NPs inside the cell, resulting in the least susceptibility. This stopped the bacterium



A B C D E

from dying

Fig. 4 Zone of inhibition on a nutrient agar plate caused by ZnO-NPs produced by an *E. coli* extract. *Salmonella sp*., *Klebsiella sp*., *Staphylococcus aureus*, *Bacillus cereus*, and *E. coli* were all evaluated using zinc nanoparticles (ZnO-NPs), in addition to tests against *Staphylococcus aureus*, *Salmonella sp*., and *Bacillus cereus*.

4.2. Characterization

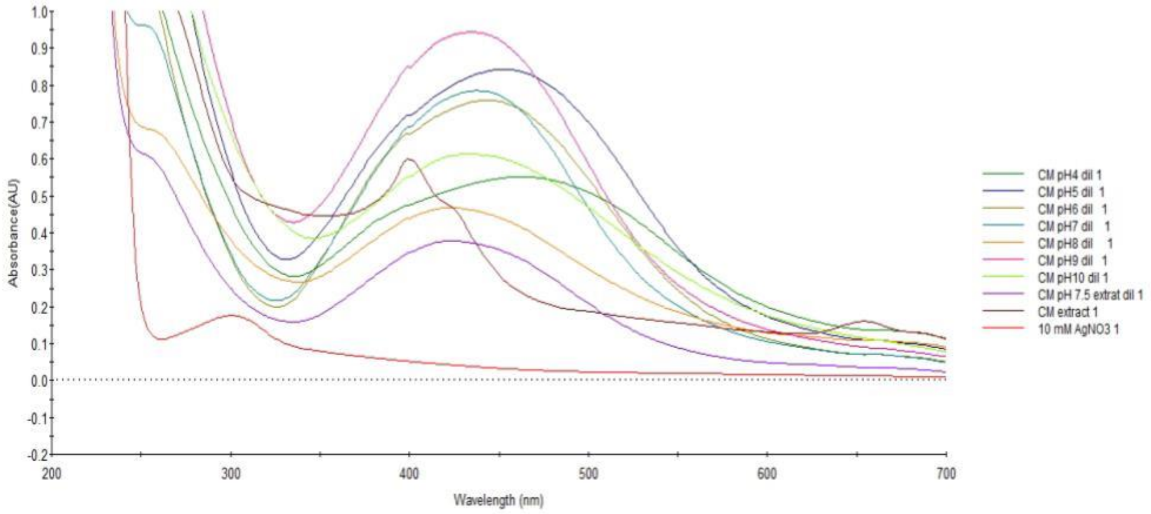
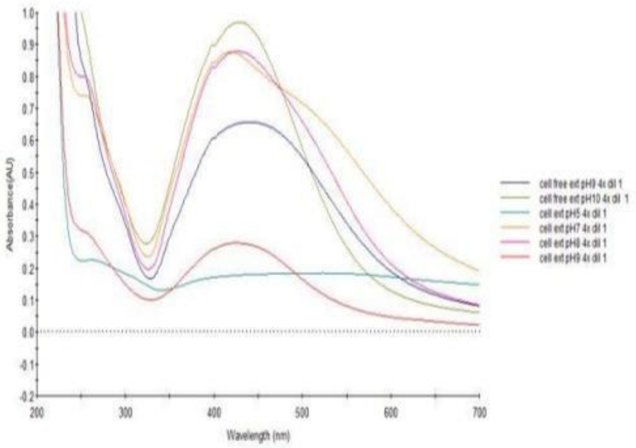
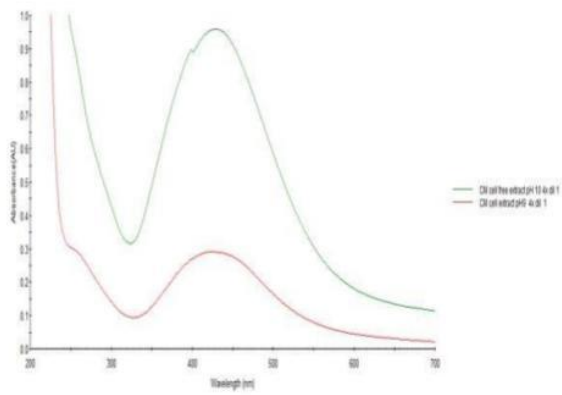


Fig. 5 Absorbance plot of Zn-NPs at 24 hours of incubation



Absorbance scan plot of synthesized Ag-NPs after 24 hours of incubation



Absorbance scan plot of synthesized Ag-NPs after 120 hours of incubation

Fig. 6 Absorbance plot at pH -9 & 10 for 24 and 120 hours incubation in light condition.

The reduced form of the metal ion and the production of the nanoparticle with a peak at 426 nm are both confirmed by the UV-Vis spectroscopic investigation, which also reveals the Plasmon resonance property.Due to their tiny size, ZnO nanoparticles used to discern colours in colloidal solution. Zinc colloids have sharp band that were seen at 426 nm, demonstrating how effectively the bacterial cell extract reduces the zinc ion. There are electron clouds on the surface of nanoparticles that may oscillate and absorb electromagnetic radiation at a specific energy, which corresponds to photons of wavelength 426 nm. Surface Plasmon resonance is the term for this resonance.

4.3 Heavy metals remediation

4.3.1 Nickel remediation

Removal efficiency is a temperature dependent and maximum removal was occurred at temperature 90°C.

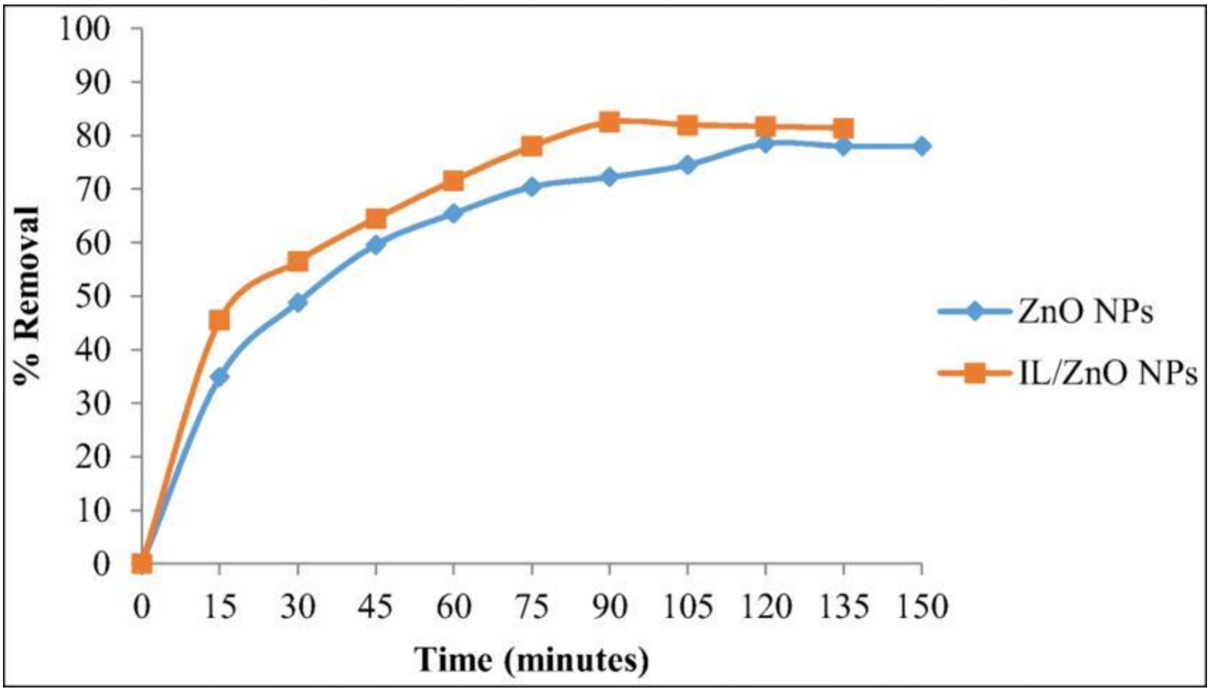


Fig. 7 Time Contact with Nickel removal

CHAPTER 5

CONCLUSION

E. coli bacteria cell extract was used to successfully create zinc oxide nanoparticles utilizing the biological green synthesis method. The reduced form of the metal ion and the nanoparticle production are both confirmed by the UV-Vis spectroscopic investigation at peak of 426, which also reveals the Plasmon resonance property. Due to their tiny size, ZnO nanoparticles include distinctive colour in colloidal solution. Zinc colloids with sharp band that were visible at 426 nm demonstrate how well zinc metal decreases bacterial cell extract.

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I, **Manesh**, **[2k21/MSCBIO/23]**, final semester student of M.Sc. Biotechnology, hereby declare that the project Dissertation titled "**Green synthesis of Zinc oxide nanoparticle by microbes and their role in heavy metals remediation**" which is submitted by me to the Development of Biotechnology, Delhi Technological University, Delhi in partial fulfillment of the requirement for the award of the degree of Master of Technology, is original and not copied from any source without proper citation. This work has not previously formed the basis for the award of any Degree, Diploma Associateship, Fellowship or other similar title or recognition.

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I, hereby certify that the Project Dissertation titled, "**Green synthesis of Zinc oxide nanoparticle by microbes and their role in heavy metals remediation**" which is submitted by, **Manesh [2K21/MSCBIO/23]**, Department of Biotechnology, Delhi Technological University, Delhi in partial fulfillment of the requirement for the award of the degree of Master of Science, is a record of the project work carried out by the student under my supervision. To the best of my knowledge this work has not been submitted in part or full for any Degree or Diploma to this University or elsewhere.

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Prof. Pravir Kumar
Head of Department

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Manesh

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